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Supplemental Material

Neurotoxicity of the Parkinson's Disease-Associated Pesticide Ziram Is Synuclein-Dependent in Zebrafish Embryos

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Figure S3: Ziram treatment results in reduction of TH-1 levels. Using Western blot analysis, the effect of ziram (50nM) on TH-1 levels was investigated. A 63% reduction in TH-1 was observed for embryos treated with 50nM ziram vs. controls (n=4; *p-value = .03 using two-tailed T test). Bars represent standard error of the mean.

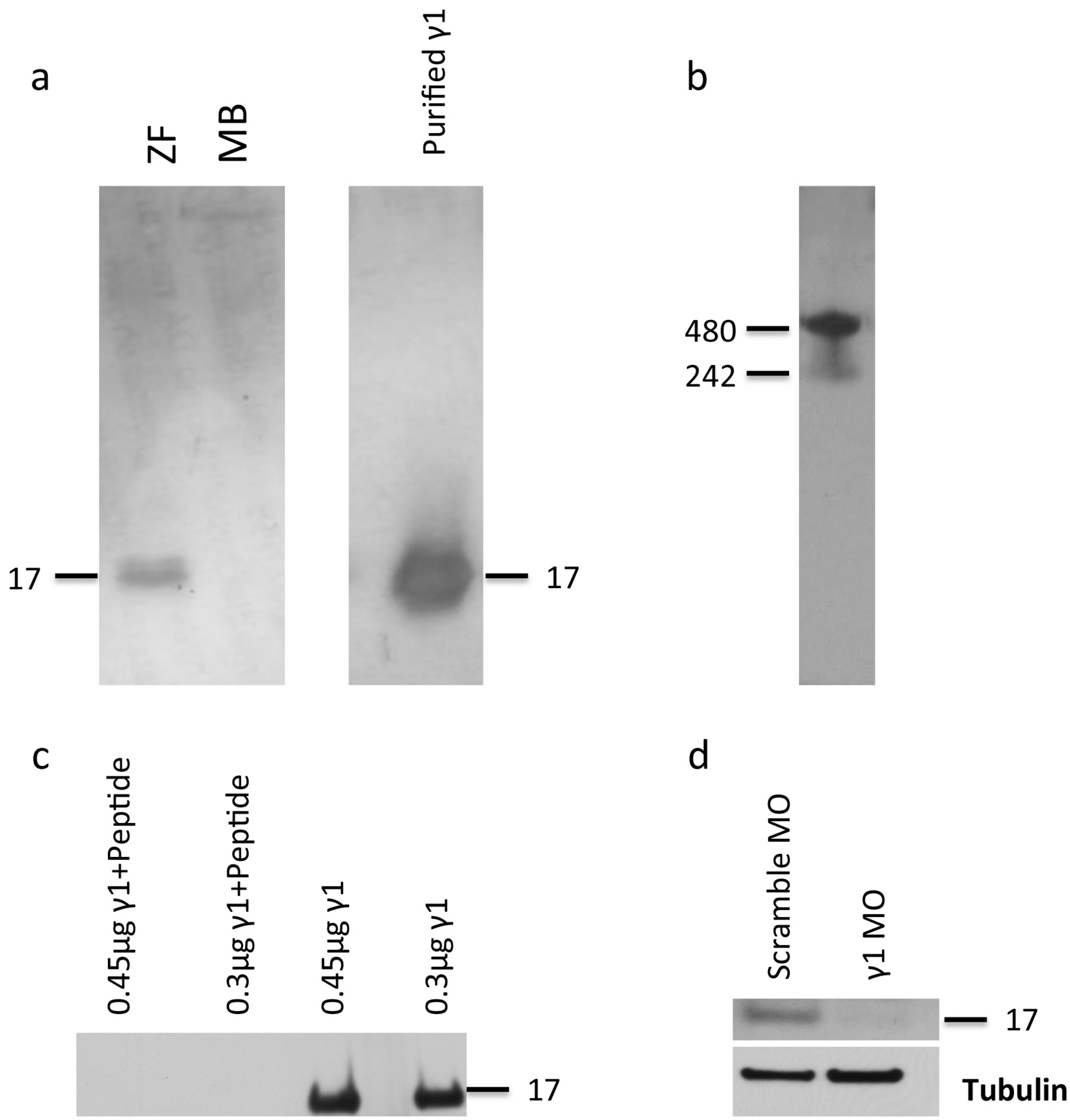


Figure S1: ZF γ1 antibody is specific for ZF. Using denaturing conditions/SDS PAGE, a band for γ1 was detected at 17 kDa for ZF adult brain (ZF) and purified ZF γ1 (a). No ZF γ1 signal was detected for mouse brain (MB). Using non-denaturing conditions/Native PAGE, a major band for ZF γ1 was detected at 480 kDa and a minor band at 242 kDa (b). Peptide preincubated with the ZF γ1 antibody is shown for SDS PAGE. No band for ZF γ1 was detected after preincubation of γ1 antibody with γ1 peptide (c). ZF γ1 MO reduced protein levels of ZF γ1 as compared to scramble MO as determined by SDS PAGE (d).

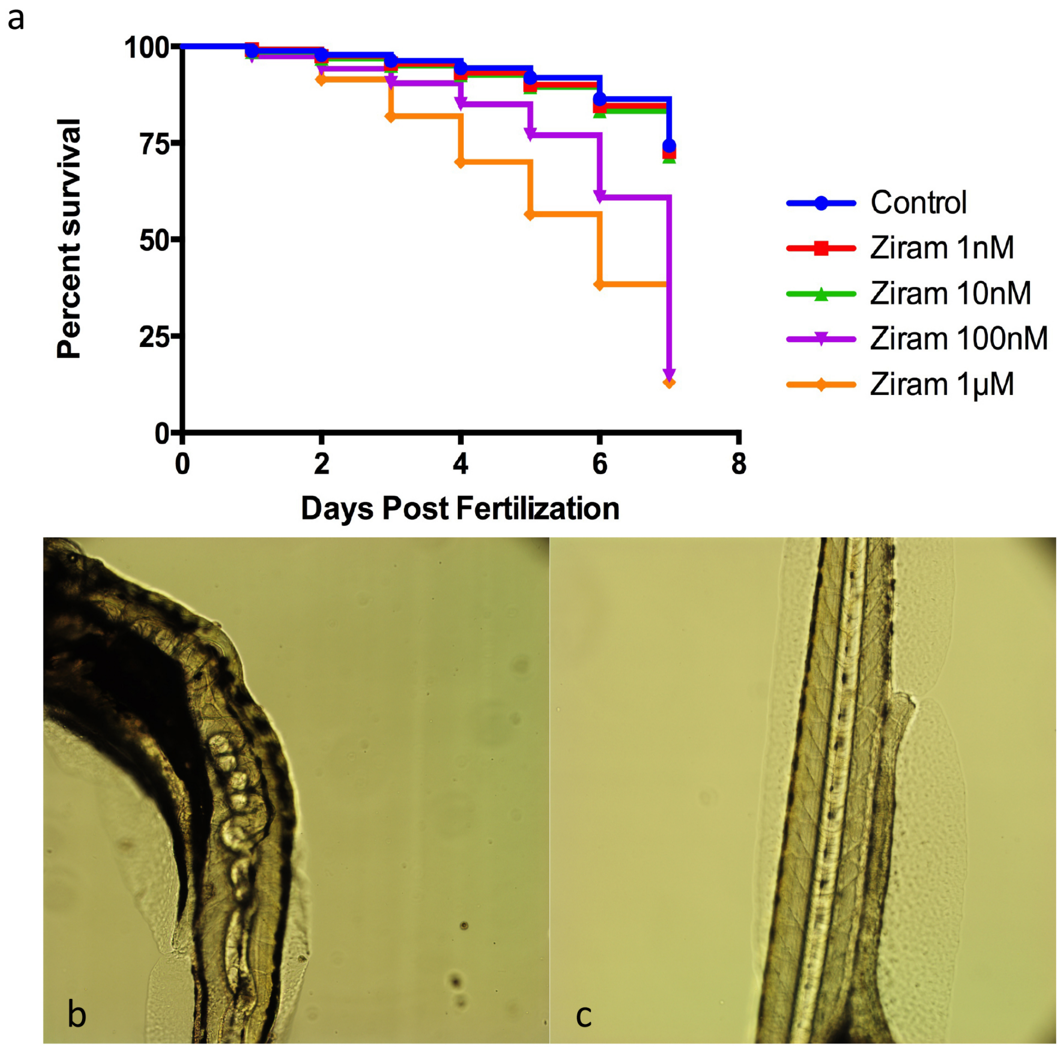
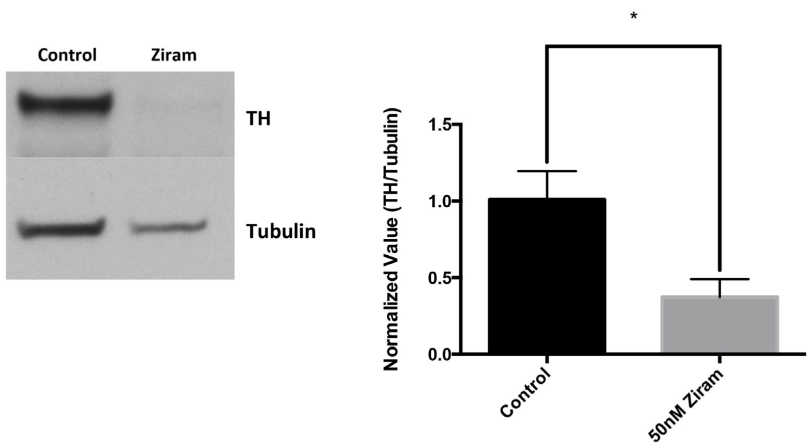


Figure S2: **Ziram is toxic in a concentration-dependent manner and causes notochord distortion.** A significant change in toxicity was observed for embryos treated (24hpf) with 100nM and 1μM ziram by day 7 (n = 50, $p<.0001$) (a). ZF embryos treated with 50 nM ziram at 5 hpf, were found to have a shorter body axis, pericardial edema, and notochord distortion (b) as compared to vehicle treated ZF (c). $p<.0001$ Log-Rank test.



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